



WATER RESOURCES RESEARCH GRANT PROPOSAL

Title: Evaluation and Management of *Cryptosporidium parvum* and Phosphorus Contributions in the Town Brook Watershed: Laboratory Experiment

Focus Categories: NPP, AG, NUT, ECL, SW

Descriptors: Solute transport, Phosphorus, *Cryptosporidium parvum*, Nonpoint pollution, Livestock agriculture, Best management practices

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Critical Water Problem:

The USEPA's Surface Water Treatment Rule generally requires filtration to remove pathogenic microorganisms from drinking water supplies. New York City (NYC) was granted an exemption from the filtration rule, providing that an acceptable watershed program plan and protective measures can be achieved. A panel of scientists convened by the National Research Council (NRC) recently carried out an independent scientific review of the completed, comprehensive NYC Watershed Program's plan. The NRC

panel recommended that NYC put its highest priority and resources on improved methods for detecting pathogens, understanding pathogen transport and fate, and demonstrating that best management practices (BMPs) will remove pathogens (NRC, 1999). Since *Cryptosporidium parvum* (*C. parvum*) oocysts are very resistant to chlorination and have multiple infection routes between humans and animals, a major focus of this research will be on their detection, transport, and on BMP methods to remove them. Phosphorus (P) removal from the NYC water supply has received the most attention to date, since P accelerates the growth of algae in streams and reservoirs. The growth and decay of algae produces dissolved organic compounds that have the potential to form toxic byproducts when water is chlorinated.

Expected Benefits

Although significant efforts are already underway through the Watershed Agricultural Program (WAP) to plan and implement BMPs on farms in the NYC watershed, uncertainty regarding the effectiveness of these practices to improving water quality is still a major concern. The basis of concern is that the predominant shallow, glacial till soils in the watershed may not be effective filters of subsurface transported contaminants, an underlying premise for many of the BMPs used to reduce surface runoff. This proposal offers a systematic approach, linking the potential transport phenomena of *C. parvum* with those of P for these shallow soils, verifying and collaborating on some intensive field and watershed scale monitoring, and developing improved models that should facilitate pathogen and nutrient planning and implementation efforts. The proposed work will involve collaborators from the College of Agriculture and Life Sciences at Cornell University in conjunction and cooperation with numerous NYC watershed cooperators including USDA-ARS scientists, USGS, the Watershed Agricultural Council, and cooperating farmers. Together with research proposed by USDA-ARS in the watershed, this project will be an important scientific component to the NYC watershed and should serve to enhance the understanding of *C. parvum* and P transport through soils typical of the region in order to improve the reliability of BMP recommendations to improve water quality.

Nature, Scope, and Objectives

Practical considerations dictate that only a limited number of BMPs can be implemented on a given field site. Consequently, a BMP should be multi-functional and control more than one priority pollutant. Since pathogens and manure P are typically transported as colloids, our hypothesis is that BMPs that are effective in controlling manure P may also be effective in controlling pathogen transport. Because of the highly infective nature of *C. parvum*, investigations with manure will be limited to the laboratory where relationships between *C. parvum* and P can be developed and evaluated. These relationships will be verified and correlated to field monitoring efforts. Based on these considerations and limitations, the objective of this proposal is to investigate process-based factors of *C. parvum* and P transport at the laboratory scale. The research result will then be used (if additional funding can be obtained) to 1) to evaluate P transport for various BMPs on field plots, 2) to conduct field/subwatershed-scale studies for the

determination of load factors, and 3) to develop and validate models for improved planning.

Methods, Procedures, and Facilities

Laboratory experiments under controlled environmental conditions can provide insight into the processes controlling P mobilization and P and pathogen transport, as well as the potential effectiveness of BMPs under field conditions. Recently spread manure is potentially an important source of pathogen and P, and it is important to examine the effectiveness of BMPs in reducing these contributions. We will investigate the effects of manure composting, manure incorporation, spreading manure away from Hydrologically Active Areas (HAAs) and the winter spreading of manure. Manure for laboratory tests will be collected from calves (which are most susceptible to infection with *C. parvum*) at the Post Farm in the Town Brook Watershed (TBW). The Post Farm will also be the source for the composted manure. The manure will be applied in these investigations at rates that would meet the agronomic N requirements for corn.

C. parvum oocyst transport through undisturbed soil columns will be correlated with the movement of dissolved constituents, P, and blue dye tracer under simulated rainfall in the laboratory. Based on our extensive experience with the preferential flow characteristics, we anticipate that this component will significantly improve our understanding of oocyst and P transport and retardation in subsurface flow, which should improve modeling of oocyst and P transport.

Soils in the TBW typically have a restrictive layer within 60 cm from the surface. Consequently, the subsurface flow of water is principally parallel to this layer. In the laboratory, undisturbed mini-hillsides with intact macropore cracks, wormholes and root channels will be simulated using undisturbed “horizontal” cores 20 cm wide, 90 cm long (downslope dimension), and 40 cm deep. These undisturbed soil blocks will be extracted from fields in the TBW. Soil cores (5 cm dia., 40 cm deep) will also be obtained adjacent to the excavations. These cores will be dissected in 5 cm depth increments to determine the background P levels by Morgan, Mehlich-3, water- and CaCl_2 -extractable P procedures.

Once in the laboratory, the soil blocks will be placed in a controlled environment room with a drain system to collect percolate. A rainfall simulator will apply water (using simulated rainwater chemistry) uniformly at low application rates. Pulses of a 25 mmol/L CaCl_2 tracer solution will also be used. Composited runoff and subsurface flow samples will be tested for *C. parvum*, P, N, and chloride. A higher frequency of sampling will be used during surface runoff and breakthrough. All collected samples will be refrigerated at 4° C, and analyzed within 24 hours.

Specific experiments to be operated include:

Composting Treatment: In this experiment, the *C. parvum* and P concentrations in runoff and interflow will be compared between composted and fresh manure as a

function of time after spreading for different environmental conditions. Composted and fresh calf manure will be surface-applied at an agronomic rate. Rainfall rates of 0.75 and 3 cm/day will be tested at temperatures of 5 and 15 C for 60 to 90 days, at which time P concentrations should have reached background levels.

Manure Incorporation: It has long been held that prompt incorporation of manure reduces the loss of N as well as pathogens and P in surface runoff. Conversely, incorporation may increase pathogen and P concentration in the interflow but little research has been done in this area. This investigation will simulate chisel plow incorporation of manure, using duplicate pairs of soil blocks and a rainfall rate of 1 cm/day at room temperature.

HAAs Avoidance: This experiment will investigate the effects of spreading on saturated (defined as HAA) and unsaturated soil conditions on pathogen and P transport. Two soil blocks will be allowed to drain freely (unsaturated condition), and two will have restricted subsurface drainage to induce saturation and overland flow. The manure will be surface-applied at room temperature. Rainfall will be applied at 1 cm/day for 60 to 90 days. Both *C. parvum* and P will be determined in the surface and subsurface flow.

Winter Spreading of Manure: The winter spreading of manure will be investigated by placing the soil blocks outdoors to expose them to the conditions of an upstate NY winter. Eight soil blocks will be used, with manure applied to two soil columns beginning in January, two columns in February, and two columns each in March and April. The interflow and surface runoff will be measured from any snow, rain, or snowmelt events. The soil blocks will be instrumented with thermocouples at 10 cm depth intervals in order to measure the temperature and frost front in the soil columns. In this treatment, no manure containing viable *C. parvum* oocysts will be applied.

C. parvum oocyst concentrations in the water and manure samples will be determined as per Anguish and Ghiorse (1997), who added a fluorescent antibody detection step to the method of Campbell et al. (1992). Analysis of water for P, N, and chloride will be done utilizing standard methods for the analysis of water and wastewater (Eaton et al., 1995), using Dionex DX-100 ion chromatograph and a Buchler digital chloridometer. Soil block experiments (except for the winter spreading treatment) will be conducted in an isolated room, with proper disinfection protocols observed. Wastes will be disinfected with Roccol-D (Winthrop Veterinary) and soils will be autoclaved at the conclusion of these experiments.

Measured outflow concentrations and rates, column areas, sample time intervals, the initial concentration in the manure, and the volume of water applied to the column will be used to determine breakthrough curves, which consist of relative concentrations (C/C_0) vs. cumulative rainfall applied or flow collected. The flux of chloride (non-retained anion) will be compared to the flux of *C. parvum* and P to determine their transport and retardation in surface and subsurface flow. Collection efficiency mass balances will be calculated by comparing application with analyte recoveries.

The results of these process-based studies will be incorporated into models which, when complete, can be used to evaluate effectiveness of BMPs in the more generic watershed context. Due to the temporal and spatial variability of pathogen sources (and the small amount of pathogens needed for human infection) risk-based factors will be included.

Related Work

Cryptosporidium parvum

Research on microbial transport through soil has largely concentrated on bacteria and viruses, which have been found at depths well below the surface and exhibit both vertical and horizontal movement. Cryptosporidiosis resulting from sewage contamination of a well was reported by D'Antonio et al. (1985) who speculated that *C. parvum* oocysts percolated through the soil and entered the well. Since *C. parvum* oocysts (4 to 6 μm in diameter) are somewhat larger than bacteria cells (generally 0.3 to 2 μm), and considerably larger than viruses (25 to 350 nm), this subsurface transport of oocysts is generally unexpected but obviously possible. More recent experiments by Mawdsley et al. (1996) and Brush (1999) show that *C. parvum* oocyst transport is similar to that of bacteria and viruses. The degree and rate of movement of bacteria and viruses in homogeneous soils is greater in coarse particles such as sand, as opposed to fine textured clay soils. The presence of macropores in naturally structured soil may exacerbate the transport of microbes via preferential flow (Madsen and Alexander, 1982; Darnault et al., 1999). The movement of *C. parvum* in preferential flow paths is one of the basic questions which this proposal will address.

Cryptosporidium may adsorb to soil during transport. Hydrophobicity, the presence of extracellular polysaccharides, and the net surface charges are factors that control adsorption. Chemical factors most critical in particle adsorption are solution ionic strength and pH, and the soil organic C content (McCaulou et al., 1995). The retention of viruses in soil is primarily due to their small size and surface properties. Virus adsorption varies greatly depending on the type of soil, the amount of organic matter, the water content, soil pH, salt content of the soil solution, flow rate, and the physical and chemical nature of the viruses. However, little is known regarding the adsorption properties of *C. parvum* and how *C. parvum* is transported in the landscape. Another unknown is what BMPs are effective barriers to *C. parvum* transport at the field and watershed scales. Some effective BMPs to reduce *C. parvum* transport may be manure composting and precision spreading away from areas that produce runoff.

Phosphorus

P is directly linked to algae growth and accelerated eutrophication of surface waters. Eutrophic conditions lead to enhanced algal growth. When the algae decay, dissolved organics are produced which can be transformed into potentially carcinogenic disinfection byproducts when the water is chlorinated. As a result, the control of P in the NYC watershed is also of critical importance.

Various studies have shown that a small number of P sources areally distributed within the watershed can contribute a significant share of total P load (Sharpley et al., 1993; Gburek et al., 1996; and Pionke et al., 1997). Cornell researchers in the Department of Agricultural and Biological Engineering have developed an analytical framework for classifying critical source-areas by their runoff potential. Three different types of critical areas have been identified: 1) shallow, sloping lands (Frankenberger et al., 1999), 2) barnyards (Kellog and Lander, 1999), and 3) flood plains (Weiler et al., 1999). Each of these areas is governed by different hydrological properties that create different patterns of P contribution. Accordingly, HAAs refer to any area or field that has, on average, greater than 30 percent probability of runoff.

Phosphorus is transported primarily via surface runoff in soluble and particulate form. However, subsurface leaching of SP may also be important in the NYC watershed. Particulate P transport is usually associated with surface erosion. Subsurface leaching of SP may occur rapidly through shallow soils (Scott et al., 1998) or more slowly where leaching reaches a deeper ground water table. Particulate P is often the major portion (75 to 90 percent) of total P (TP) transported from cultivated lands, while SP is the major contributor from non-cultivated lands. The relative speciation of P is important to management efforts because of varying reactivity and bio-availability to algae. A major effort on determining soil P dynamics was conducted by Kleinman et al. (1999) in the TBW from 1997-1999. It was determined that 50 kg/ha Morgan soil test P was found to be a critical level for the soils that are characteristic of the TBW. In addition, changes in soil P fractions as a function of manure application rate and soil type were measured (Kleinman, 1999). This background monitoring and the partnerships which have been established will facilitate the ability to carry out the objectives identified in this proposal.

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